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TECHNICAL MANUSCRIPT 493

ADVANCES IN LARGE-VOLUME AIR SAMPLING

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Herbert M. Decker Lee M. Buchanan David E. Frisque Melvin E. Filler Charles M. Dahlgren

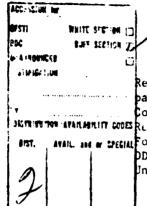
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DEPARTMENT OF THE ARMY Fort Detrick Frederick, Maryland 21701

TECHNICAL MANUSCRIPT 493

ADVANCES IN LARGE-VOLUME AIR SAMPLING

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December 1968

ABSTRACT

A review of the most recent advances in the field of large-volume air sampling is provided. The electrostatic precipitator, the multi-slit, and the liquid-scrubber large-volume air samplers are discussed and data are presented on their effectiveness in recovering aerosols of Serratia marcescens and B. subtilis var. niger. The authors conclude that much progress has been made recently, and that air-sampling equipment is now readily available that will remove a high percentage of ambient microorganisms in viable form from a large volume of air, and in addition will concentrate them into a small volume of liquid.

I. INTRODUCTION

The Engineering and Sanitation Section of the American Public Health Association suggested that we summarize the current state-of-the-art in the field of large-volume air sampling.

There is an obvious need for air-sampling devices that will recover a high percentage of ambient microorganisms in viable form from a large volume of air and, in addition, will concentrate them into a small volume of liquid. This requirement often faces scientific investigators studying the airborne transmission of disease in research laboratories and hospital rooms. Studies of laminar flow as a biological barrier also require sampling systems that collect large volumes of air to assure the effectiveness of the system.

II. AVAILABLE LARGE-VOLUME AIR SAMPLERS

A. DESCRIPTION

During the past few years, as an outgrowth of a Fort Detrick development contract, large-volume air samplers capable of operating at sampling rates up to 15,000 liters per minute have become commercially available. At least two firms stock several types of large-volume air samplers that operate on the principle of electrostatic precipitation of airborne particles onto a wetted rotating disc. These samplers come in two ranges of air flow rates, 300 to 1,200 liters per minute and 2,500 to 15,000 liters per minute. For ease of calculation, the samplers are normally operated at 1,000 or 10,000 liters per minute.

The air enters the inlet of the LVAS* sampler (Fig. 1) and passes through a corona discharge at the center of the disc, which negatively charges the particles. The air then passes between the high-voltage plate and the rotating porcelain collection disc. The particles negatively charged by the corona are attracted to the collection disc, which is at ground potential, and the air is drawn through the sampler to the exhaust by a centrifugal blower.

The organisms attracted to the rotating collection disc are concentrated in a liquid that flows over the disc. This liquid is drawn from a reservoir to the center of the collection disc by a peristaltic pump and is spr2ad uniformly and continuously by an adjustable monofilament

^{*} Litton Systems, Inc.

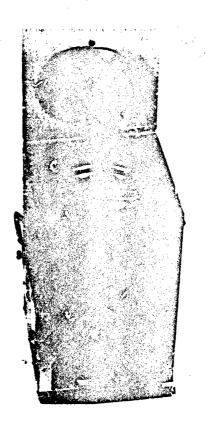


FIGURE 1. Litton Systems, Inc., Electrostatic Precipitator Large-Volume Air Sampler (LVAS).

wiper over the porcelain surface of the collection disc. The liquid flows into a groove at the periphery of the disc and is then picked up by a suction probe and pumped into an effluent reservoir.

Initial studies on large-volume air samplers were directed toward the collection of the total number of airborne organisms, both viable and nonviable. Tests on the early research models of the electrostatic precipitator samplers indicated organisms from 10,000 liters of air could be concentrated into 10 ml of a collection liquid containing 90% glycerine and 10% water. The absolute collection efficiency approached 80 to 90%. As more became known about this type of sampler, it became a useful tool for the recovery of viable airborne microorganisms when a more compatible collection fluid was used to collect the lable organism. For example, the Public Health Service has used the larger air sampler to collect viable rabies virus from the air in a bat cave in Texas.* This is the first time the presence of this virus in a natural aerosol form has been

^{*} Winkler, W.G. 1968. Airborne rabies virus isolation. Bull. Wildlife Dis. Ass. 4:37-40.

demonstrated. Studies also have been conducted by the Army Medical Department and Fort Detrick to determine the presence of the upper respiratory pathogens, meningococcus and adenovirus type 4, in aerosol form in barracks where recruits were housed.*,**

B. SAMPLER EVALUATION

During the past year, one of the latest commercial versions of the 1,000-liter-per-minute large-volume electrostatic precipitator sampler was tested with separate aerosols of <u>Bacillus subtilis</u> var. <u>niger</u> spores and <u>Serratia marcescens</u> cells. Both aerosols consisted mainly of particles with a 1-micron number median diameter and containing a single cell.

The test aerosols were produced by continuously spraying, individually, suspensions (10^7 and 10^9 cells per ml) of <u>B. subtilis</u> and <u>S. marcescens</u> from a Vaponefrin nebulizer. The aerosol was directed into an environmental chamber (Fig. 2), where it was thoroughly mixed with filtered air, then circulated through a sampling duct, and finally returned to the chamber through a closed duct system.

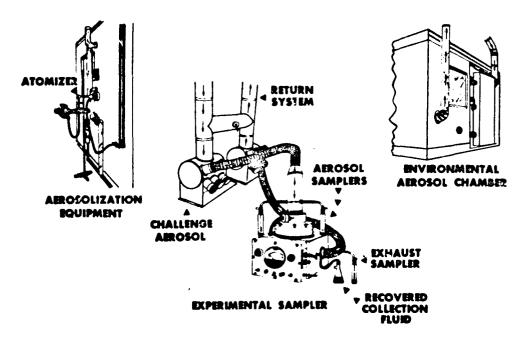


FIGURE 2. Air Sampler Test Equipment.

^{*} Artenstein, M.S.; Miller, W.S. 1966. Air sampling for respiratory disease agents in Army recruits. Bacteriol. Rev. 30:571-572.

^{**} Artenstein, M.S.; Miller, W.S.; Lamson, T.H.; Brandt, B.L. 1968.
Large-volume air sampling for meningococci and adenoviruses. Amer. J.
Epidemiol. 87:567-577.

After a 30-minute equilibration period, the aerosol was drawn from the sampling duct through the sampler and then exhausted to the return duct system. Under these conditions, an aerosol concentration of 1,100 to 1,500 B. subtilis spores and 10,000 to 15,000 S. marcescens organisms per liter of air was generated. In all tests on the electrostatic precipitator sampler, normal growth tissue culture medium containing 0 2% Methocei* (a methylcellulose product with a viscosity of 15 centipoise) was used as the collection liquid in the sampler. This medium was selected on the basis that it would serve as a universal collection medium for the viable recovery of bacteria, viruses, fungi, and rickettsiae. The components are Hanks balanced salt solution and lactalbumin hydrolysate. Separate experiments conducted previously showed that the addition of 0.2% Methodel indicased the viscosity and lowered the surface tension of normal growth tissue culture medium and resulted in higher recoveries of S. marcescens and B. subtilis. The 1,000-liter-per-minute sampler requires a liquid flow rate of approximately 5 to 6 ml per minute; the 10,000-liter-per-minute sampler requires tenfold more. Experience has shown that it may be necessary to vary the liquid flow rate, depending upon the viscosity and surface tension of the collection medium and the ambient temperature and relative humidity.

All-glass impingers (Acl-30) were used for this evaluation. They were operated at 12.5 liters per minute for each 5-minute test period; two were placed upstream of the sampler to determine the challenge aerosol concentration, and one was placed downstream to determine the effluent (or slippage) concentration.

The collection liquids for the ACI-30 samplers were distilled water (20 ml) for sampling <u>B</u>. <u>subtilis</u> aerosols and normal growth tissue culture medium (20 ml) containing 0.06% Dow Corning Antifcam A for sampling <u>S</u>. <u>marcescens</u> aerosols. The different collection fluids were selected on the basis of maximum collection efficiency, physiological compatibility with the organism, and physical compatibility with the sampler.

Immediately after each test the appropriate serial dilutions were made in distilled water for <u>B. subtilis</u> and in normal growth tissue culture medium for <u>S. marcescens</u>. The collection liquids were then assayed on streak places of Facto tryptose agar. Tolonies were counted after 16 to 20 hours' incubation at 37 C.

C. RESULTS OF SAMPLER E ALIATION

Forty 5-minute tests were performed on the electrostatic precipitator high-volume sampler. The collection efficiency (based on the AGE sampler as 100% efficient) averaged 95% for S. marcascens and 74% for B. subtilis. Specific studies have shown that there is an apparent interaction between gram-positive spore formers and normal growth tissue culture medium that does not occur with S. marcascens and several other gram-negative organisms studied.

^{*} The Dow Chemical Lo.

Experience has shown that large-volume air samplers are far more sensitive and complex than the commonly used samplers such as the agar slit sampler and the all-glass impinger. Until recently it was necessary to caution those who used electrostatic precipitator samplers to ascertain that (i) they were operating at the voltage recommended, (ii) the surface of the rotating disc was completely wet, and (iii) the proper distance was maintained between the sampling head and the rotating disc. In other words, one could not assign an untrained employee to operate the sampler. Recently, considerable engineering and development work, with subsequent retrofitting, has been done on these types of samplers, and we now are approaching a period where they can be left unattended and there is still reasonable assurance that the sampler is running properly.

III. RESEARCH INVESTIGATIONS

Because of the need for a large-volume air sampler that is both reliable and simple, this laboratory has sponsored research both in-house and under contract to explore the possibility of developing improved devices. Areas of investigation include samplers operating on the principles of:

- 1) Electrostatic precipitation in the field produced by a space charge;
- 2) Electrostatic precipitation in the field produced by charged parallel plates;
- 3) Electrostatic precipitation of particles onto oppositely charged liquid droplets;
 - 4) Air washing with subsequent removal by a cyclone;
 - 5) Inertial impaction by a rapidly moving surface; and
 - 6) Inertial impingement onto a wetted surface.

Of all these approaches, the principles of inertial impingement on a wetted surface and of air washing with subsequent collection in a cyclone appear most promising. Of particular interest to the investigators at Fort Detrick is a very simple sampler called the multi-slit impinger. The principle of its operation was adapted in part from the single-slit agar sampler we developed several years ago for the direct impaction of bacterial particles onto a rotating agar plate. The agar slit sampler (Fig. 3) is now available in a commercial version and has been widely adopted and used in public health laboratories as well as in our own research.

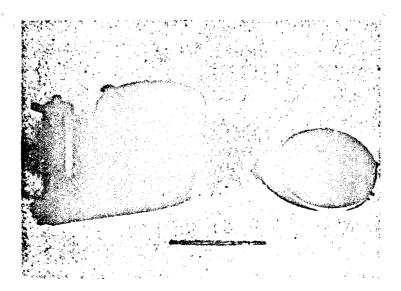


FIGURE 3. Fort Detrick Slit Sampler.

The initial multi-slit impinger sampler (Fig. 4) developed in this laboratory in mid-1967 operates on the principle of inertial impingement of airborne particles into a liquid film maintained on the surface of a rotating disc. Air is drawn into the sampler at 500 liters per minute through eight small rectangular slits located very near the surface of the disc. The collection liquid is pumped to the center of the rotating disc through a small-diameter stainless steel tube suspended above and across the diameter of the disc. This tube also spreads the liquid uniformly over the disc surface. The high-velocity air jets directed against the liquid film impinge the airborne particles onto the liquid. The particle-laden-liquid then flows across the surface of the disc and is removed by a hollow plastic scraper at the rim of the disc that allows the liquid to pass into a collection tube, where it is removed by vacuum into a container.

This sampler (Fig. 5) recently was improved under a contact program with Environmental Research Corporation to determine optimum sampler design and operating conditions. Such parameters as slit width, slit-to-disc distance, and liquid input rate were investigated. The most desirable design and operational features were selected on the basis of test data. This sampler, which is still under development, now operates at 1,000 liters per minute. Other modifications include changes to the angular placement of the slits and the collection liquid feed system. In addition, the sampler was made into a self-contained unit by incorporating a peristaltic pump to supply and remove the collection fluid, and a centrifugal blower. This sampler measures approximately 13 by 13 by 16 inches.

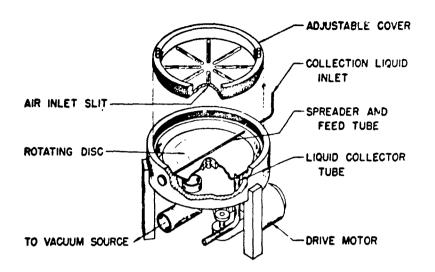


FIGURE 4. Fort Detrick Multi-Slit Sampler.

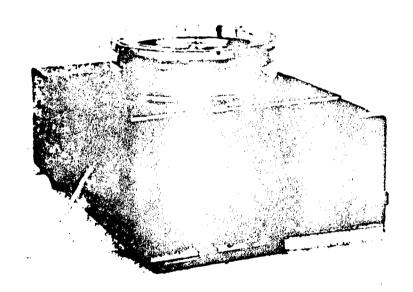


FIGURE 5. Environmental Research Corporation Multi-Slit Sampler.

A liquid-scrubber aerosol collector (Fig. 6) is being developed under a Government contract with Aerojet-General Corp. It operates at an airflow rate of 1,000 liters per minute and shows considerable promise. This sampler consists of a spray device, a collection tube, a glass coalescence tube and a cyclone mist separator containing a wash tube. The principle of operation is based on the production of a fine mist in a rapidly moving airstream, with ultimate collection of the aerosol in the mist or impaction on the elbow with subsequent washoff. The particulates in the mist are collected at the effluent end of the cyclone.

The same type of aerosol test apparatus described previously was used in twenty 5-minute tests with <u>S. marcescens</u>; the average collection efficiency of the multi-slit sampler was 90% (Table 1). For the liquid scrubber, efficiencies averaged 70%. These percentages are based on an assumed 100% efficiency for the AGI-30 sampler.

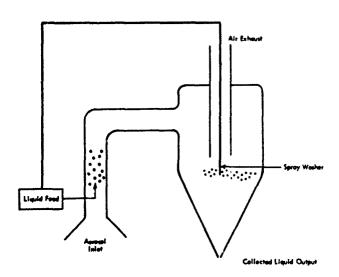


FIGURE 6. Aerojet-General Liquid-Scrubber Air Sampler.

TABLE 1. PER CENT EFFICIENCY OF LARGE-VOLUME AIR SAMPLERSAL

	Per Cent Efficiency with Indicated Test Organisms			
Type of Sampler	S. marcescens	B. subtilis var. niger		
LVAS Electrostatic Precipitator	95	74		
Environmental Research Corporation Multi-Slit	90	86		
Aerojet-General Liquid Scrubber	70	63		

a. Sampling rate - 1,000 liters per minute. Results are averages of twenty tests.

In similar tests with <u>B. subtilis</u>, the collection efficiency of the multi-slit sampler was 86% and, for the liquid scrubber, 63% (based on the AGI-30 sampler as 100%). In these tests Methocel was not used because the multi-slit sampler is equipped with a wiper and is not dependent upon a surfactant for uniform wetting. The liquid scrubber functioned better in the absence of Methocel.

b. Per cent efficiencies are based on the all-glass impinger sampler (AGI-30) as 100%.

IV. DISCUSSION

It is encouraging that considerable progress is being made in improving the methodology and sensitiveness of air sampling. One point should be stressed: all these samplers are constantly being modified and improved, and the efficiency figures reported here are subject to change. Whether a sampler is 80% efficient or only 70% is not the important thing. The importance lies in the total number of particles recovered for assay, which is a function of the sampler efficiency and the volume of air sampled. Simplicity of design, compatibility of the collecting medium for sampling the organisms, operational dependability, and cost also must be considered in the final selection of the sampler.

This brief review does signify a turning point in the field of microbiological air sampling. We are now at the threshold of sampling large volumes of air to determine the presence of microbiological flora in research laboratories, hospitals, and clean rooms.

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